

TJA-139US
10/577840
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Amendments to the Specification:

TITLE:

Please replace the title with the following amended title at page 1, line 1:

IMMUNOTHERAPEUTIC AGENT FOR THE COMBINED TREATMENT OF TUBERCULOSIS
IN ASSOCIATION WITH OTHER DRUGS

~~IMMUNOTHERAPEUTIC AGENT WHICH IS USED FOR THE COMBINED TREATMENT OF~~
~~TUBERCULOSIS TOGETHER WITH OTHER PHARMACEUTICALS~~

Please add the following new paragraph after the title at page 1, line 5:

This application is a U.S. National Phase Application of PCT International Application
No. PCT/ES2004/000482, filed October 29, 2004.

Please replace the heading at page 1, line 5 with the following new heading:

Field of invention~~Technical Field~~

Please replace the heading at page 1, line 11 with the following new heading:

Background of the invention~~Prior Art~~

Please replace the paragraph at page 1, line 29 with the following rewritten paragraph:

This prolonged treatment may induce the development of microorganisms resistant to these drugs when the treatment is not completed and, moreover, the aforementioned drugs only act when the bacillus has an active metabolism (i.e., when it is growing) but not when it has a non-active metabolism. This is a significant ~~inconvenient~~inconvenience because during tuberculosis infection bacilli coexist in both an active and a non-active metabolism phase.

Please delete the heading and paragraphs beginning at page 2, line 19 and ending at page 2, line 27:

Object of the invention

Please add the following new paragraphs at page 2, line 30:

The invention provides a method to obtain an immunotherapeutic agent containing cell wall fragments of a virulent MTB-C strain, which is useful for the combined treatment of tuberculosis in association with other drugs.

The invention further provides an immunotherapeutic agent and pharmaceutical compounds obtained using this method and the use of these compounds for treating a person with tuberculosis.

Please replace the paragraph at page 2, line 30 with the following rewritten paragraph:

The authors of this invention have discovered a method to obtain an immunotherapeutic agent that contains cell wall fragments from a virulent *Mycobacterium tuberculosis*-complex (MTB-C) strain. This is characterized in that it includes the following steps:

Please replace the paragraph at page 3, line 12 with the following rewritten paragraph:

As regards this invention, the culture must be done-maintained for a period of time of three weeks or longer, preferably between 3 and 4 weeks. The temperature of the culture is preferably maintained at between 34°C and 38°C.

Please replace the paragraph at page 3, line 15 with the following rewritten paragraph:

Following the completion of the culture, if it has been conducted in a solid phase, the plates are scrapped-scraped to obtain the colonies while avoiding media extraction (agar). Nevertheless, if the culture has been conducted in a liquid phase, the cells are concentrated and washed using conventional techniques known by a person skilled in the art (e.g., centrifugation).

Please replace the paragraph at page 3, line 20 with the following rewritten paragraph:

The homogenization of the strains is carried out in a buffered media at a neutral pH. In this invention, it is important that the homogenization is conducted in the presence of a non-ionic tensioactive compound that favors the obtaining of finely divided cell wall particles and at least partly-partial emulsions of unwanted lipidic fractions.

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Please replace the paragraph at page 3, line 26 with the following rewritten paragraph:

The homogenization may be carried out using sonication by ultrasounds, or small beads with a diameter of approximately 1 mm (e.g., of silica or silica-zirconium) together with a ~~mechanic~~mechanical homogenizer. A ~~mechanic~~mechanical homogenizer such as the BEADBEATER model of the company Biospec may be used.

Please replace the paragraph at page 3, line 32 with the following written paragraph:

The type of non-ionic tensioactive compound used is not crucial, although it is preferable to choose one from the ~~acylphenol ethoxylatalkylphenol ethoxylate~~ group and the ethoxylated sorbitan esters. It is better that the non-ionic tensioactive compound is selected from the octylphenol ~~ethoxylat~~ethoxylate compounds. Most preferably, octylphenol ~~ethoxylat~~ethoxylate with 7-8 mol of ethylene oxide are used; these may be found in the market under the name TRITON X-114, for example. The concentration of the non-ionic tensioactive compounds during homogenization ranges between 1 and 5% of the total weight homogenized.

Please replace the paragraph at page 6, line 33 with the following rewritten paragraph:

The sediment is kept, and the yellowish supernatant is ~~eliminated~~discarded.

Please replace the paragraph at page 7, line 5 with the following rewritten paragraph:

Washing and centrifugation are repeated~~, and until~~ the supernatant obtained is completely clear. After each centrifugation the supernatant~~, and~~ is discarded.

Please replace the paragraph at page 8, line 10 with the following rewritten paragraph:

The bacillary concentration, i.e., the number of viable bacilli, is determined by the incubation of ~~seriated~~serial dilutions of homogenized left lung and spleen in Middelbrook 7H11-type agar. Left lung and spleen samples were homogenized in the presence of 1 mL of PBS buffer.

Please add the following new paragraph at page 13, line 13:

Although the invention is illustrated and described herein with reference to specific embodiments, the invention is not intended to be limited to the details shown. Rather,

various modifications may be made in the details within the scope and range of equivalents
of the claims and without departing from the invention.